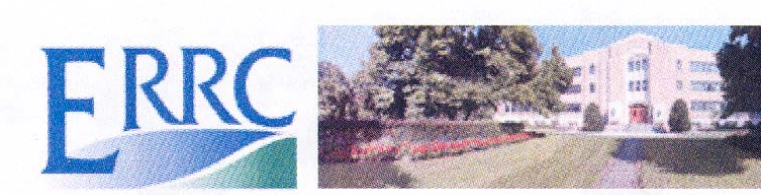

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Title: Scale-Up and Field Test of the Vacuum/Steam/Vacuum Surface Intervention Process for Poultry

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SCALE-UP AND FIELD TEST OF THE VACUUM/STEAM/VACUUM SURFACE INTERVENTION PROCESS FOR POULTRY[1]

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ABSTRACT

The Vacuum/Steam/Vacuum surface intervention pilot plant processor was scaled up to a mobile unit that can be transported to close proximity of chicken processing plants. After several modifications to the mandrel that supports the broiler carcass in the treatment chamber to minimize mechanical damage, the unit was capable of 1.1 log cfu/mL kill of inoculated Listeria innocua *and 1.4 log cfu/mL kill of inoculated* E. coli *K-12. Field tests achieved 1.4 log kill of* E. coli *and 1.2 log kill of* Campylobacter *on freshly processed chicken using 3 cycles and 138C saturated steam. But, there was extensive mechanical damage. The mandrel was modified in the Eastern Regional Research Center pilot plant to eliminate the mechanical damage. With mechanical damage eliminated, the bacteria kill was 1.1-1.5 log of inoculated* E. coli *K-12 with a total process time of 1.1 s.*

[1] Mention of a brand or firm names does not constitute an endorsement by U.S. Department of Agriculture over others of a similar nature not mentioned.

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INTRODUCTION

Processes such as blanching, cooking, canning, and pasteurizing destroy harmful bacteria on foods. However, unprocessed raw foods harbor bacteria, sometimes pathogenic bacteria. The many food recalls attest to the occurrence of bacteria slipping through the food system to product distribution.

Raw chicken frequently has pathogenic bacteria in and on the surface, for example *Salmonella* and *Campylobacter*. Although it is rare for anyone to eat raw chicken, under cooking and cross contamination from chicken can lead to food poisoning.

Treatment of these bacteria with steam can kill bacteria but will thermally damage the chicken surface. Morgan *et al.* (1996a) showed that application of steam to the surface could be accomplished with virtually no thermal damage if the process can be made sufficiently rapid. The chief deterrent to rapid treatment is the presence of a minute layer of air and water on the surface. Morgan *et al.* (1996b) developed a concept in which the interfering layers of air and water are first removed by vacuum followed by application of saturated steam. Then reapplication of vacuum cools the surface, stopping any thermal damage.

These same researchers (Morgan *et al.* 1996a) developed a machine capable of evacuating the surface, applying steam, and evacuating the surface again in less than 1 s. The process is called the vacuum/steam/vacuum (VSV) surface intervention process. The speed of the treatment is necessary to prevent thermal damage and to equal the speed of chicken process lines, nominally 70 birds per minute or greater.

Morgan *et al.* (1996b) were able to achieve 2-2.5 log bacteria kills on inoculated spots on chicken parts using the VSV process. In a side study at the request of hot dog processors, Kozempel *et al.* (2000a) found that cycling the treatment for hot dogs enhanced the bacterial killing. Apparently, the condensing steam immediately forms a resistance layer to further treatment. By cycling between vacuum and steam, the condensed steam is removed and fresh steam contacts the surface. With hot dogs, it was possible to achieve up to 5 log kill of inoculated *Listeria innocua* using 3 cycles. Although the overall kill was considerably less, cycling enhanced bacteria kill on chicken also.

When the VSV process was tested on whole broilers, there was nc statistically significant bacteria kill (Kozempel *et al.* 2000b). The difficulty treating the whole carcass was the failure of the VSV process to treat the cavity of the carcasses. The VSV process chamber was retrofitted to include a mandrel to hold the carcass and specifically treat the internal cavity (Kozempel *et al.* 2001). Unfortunately, this retrofit prototype VSV machine was not a true prototype of a commercial unit. The mandrel was locked into the product valve, therefore, it rotated to the horizontal during processing allowing the chicken carcasses to slide partially off the mandrel. Although the retrofitted mandrel

proved that the concept of a mandrel would work, the actual bacteria kill was only about 0.7-0.8 log. A new VSV processor was needed in which the mandrel was operated independently of the main valve. It was also desirable to design and build a mobile unit that could be used in the field to test at actual poultry processing plants.

A new field VSV surface intervention process machine has been designed and fabricated with a mandrel designed independent of the main valve movement. We hypothesize this unit will achieve better bacteria kills than the previous unit using freshly slaughtered and processed prechill chicken carcasses. The objective of this study was to scale-up the prototype pilot plant unit to a mobile unit and conduct a field test.

MATERIALS AND METHODS

VSV Surface Intervention Processor Mechanical Design

The surface intervention processor was designed to process chicken carcasses, specifically broilers. The performance requirements for chicken are to suspend carcasses individually on a mandrel in a chamber within a rotor; to evacuate the chamber; to inject saturated steam into the chamber; to draw vacuum on the chamber to evaporatively cool the carcass; and finally to eject the carcass into a clean environment. A prototype field unit containing one chamber and a mandrel in one rotor, was designed and constructed. Figure 1 shows the processor and Fig. 2 shows details of the product treatment section that includes the mandrel. The spherical chamber (product valve) is 254 mm in diameter.

To admit vacuum or steam into the closed chamber, two opposed 200 mm holes were bored through the ball valve stator (housing) at right angles to both the axis of rotation of the ball and to the centerline of the open chamber (carcass entry and exit ports) as shown in Fig. 2. Two platter valves are close coupled to these 200 mm ports. Each consists of a flat disk with two holes or ports rotating against an inlet header, which holds PEEK (polyetheretherketone) seals. When the disk is aligned with the ports in the inlet header, gas flows into or out of the treatment chamber. Multiple holes reduce the disk angular movement necessary for valve action and increase the cross sectional area for gas flow.

A mandrel (Fig. 3) was designed and fabricated that supports the carcass, treats the visceral cavity, and orients the carcass independently of the main valve position. A servo-actuated three-way valve applies the cavity treatment. The treatment valve common port is connected to the mandrel via a 19 mm stainless steel tube directed into the cavity. The other ports of the valve are connected to the steam and vacuum sources. A second drive rotates the 19 mm tube in the vertical plane about an axis concentric to that of the main valve. The rotating

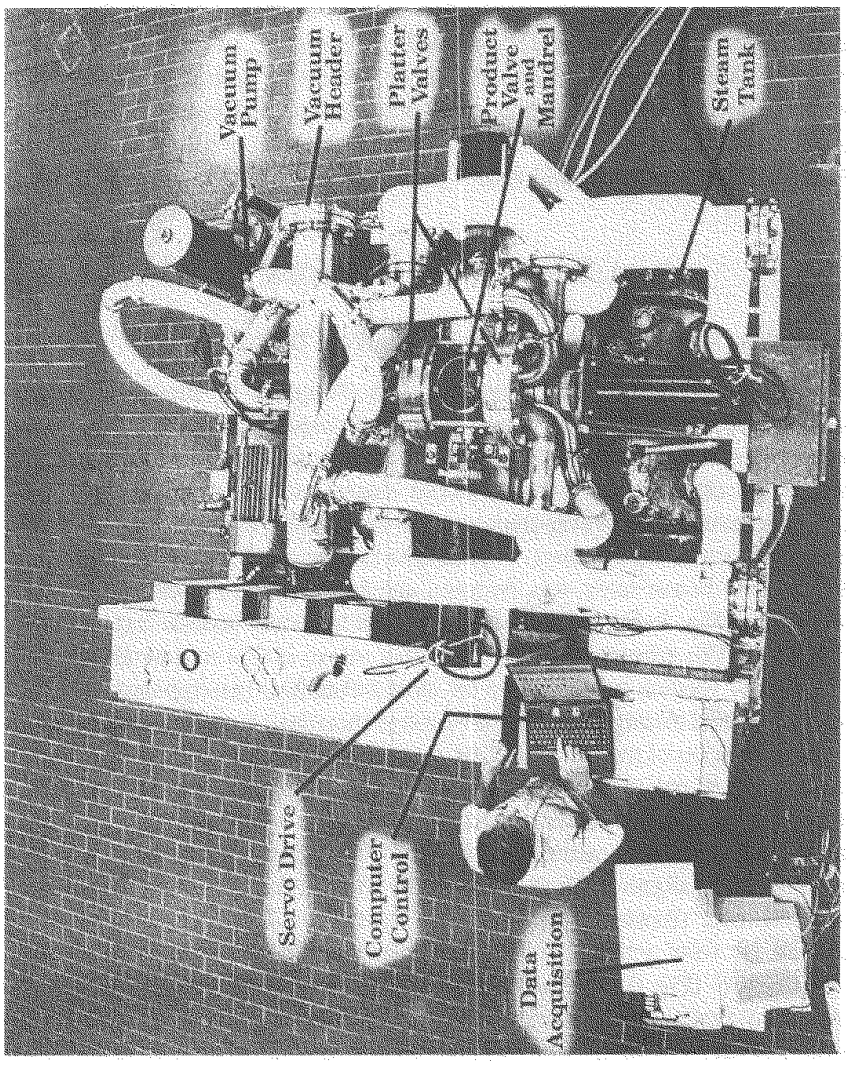

FIG. 1. PHOTOGRAPH OF THE FIELD VSV SURFACE INTERVENTION PROCESSOR, TOP VIEW

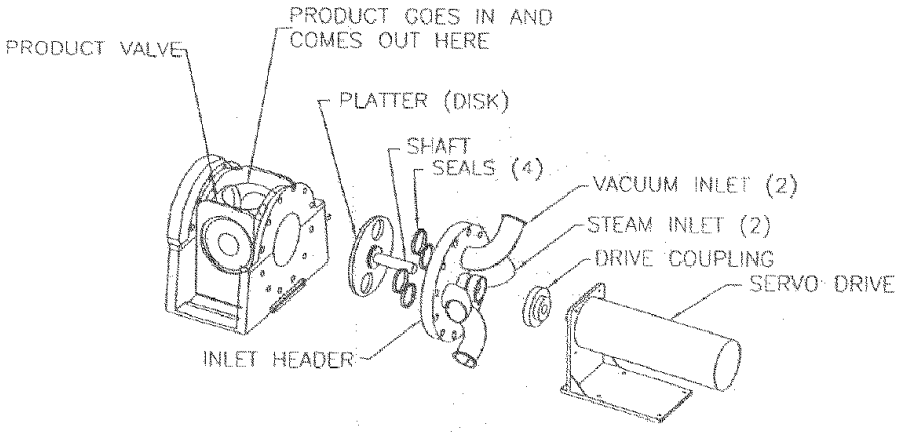

FIG. 2. EXPLODED VIEW OF ONE OF THE TWO PLATTER VALVES ON THE FIELD VSV SURFACE INTERVENTION PROCESSOR

mandrel allows flow-through handling of product and allows the product to be oriented to any position during treatment.

The mandrel, Fig. 3, is an open frame formed of three 6.4 mm diameter stainless steel rods shaped to fit the visceral cavity of a broiler carcass which is located under the breastbone and clavicles. They approximate the internal dimensions of the cavity and give lateral stability to the carcass during processing. A bottom support is included to keep the legs within the chamber and help support the carcass. The mandrel is fixed to the 19 mm treatment tube on an adjustable mount to accommodate different length carcasses. The treatment tube was also fitted with various orifice plates to support reduced flow experiments. The selected treatment tube orifice plate was 13 mm internal diameter.

Each disk and mandrel is programmed and controlled independently and moved by its own servomotor. The servos are by Allen-Bradley Co., Mayfield Valley, OH and capable of high acceleration and deceleration. The servos for the disks are model 1326AB-C4B-11, 5.6 kW, capable of 1600 rpm maximum. The servo for the mandrel is model 1326AB-B2E-11, 2.5 kW, capable of 3000 rpm maximum. The servos are direct coupled mechanically to the disks and mandrel. Operation of the servos, controlled by Graphics Motion Language (GML) software version 3.8.2, Allen-Bradley Co., Mayfield Valley, OH, controlled the vacuum and steam times. Data acquisition was by Laboratory Technologies Corp., Lab Tech Notebook version 8.04, Wilmington, MA. Sensors were by Omega Engineering, Stamford, CT. Type E thermocouples were used for temperature and Omega PX176 series sensors were used for vacuum and for steam pressure.

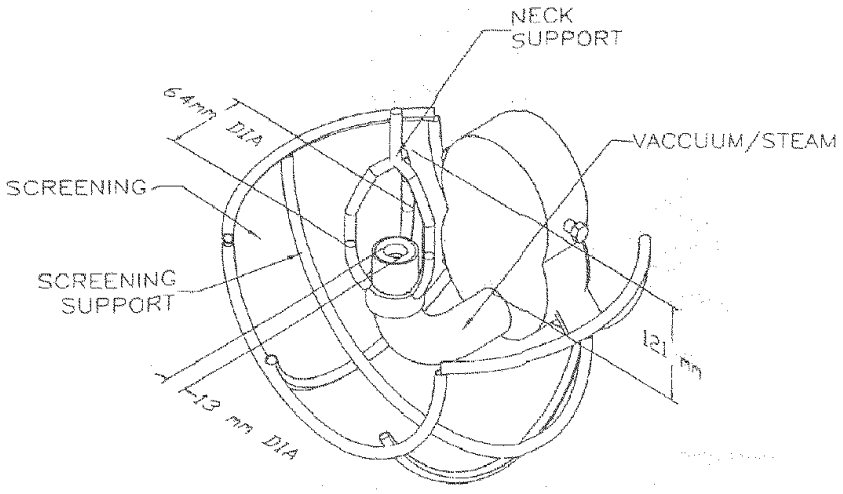

FIG. 3. THE MANDREL ASSEMBLY FOR THE FIELD VSV SURFACE INTERVENTION PROCESSOR

Vacuum was supplied by a liquid ring vacuum pump, Stokes Vacuum, model HER, Philadelphia, PA. The steam generator volume was 115 L. The stream generator contained horizontal submerged coils with no separator and 17.8 kW heaters. It was fabricated in-house and charged with water which was boiled for 30 min for deaeration. The vacuum receiver was adjusted to 7 kPa and its condenser coil cooled to 4C.

VSV Surface Intervention Processor Operation

Each sample was manually inserted into the treatment chamber of the surface intervention processor. The computer controlled ball valve was rotated, with a servo, 90° to seal the chamber from the outside atmosphere. Each servo/disk and servo/mandrel combination is called an axis of rotation. The platter valves in the main chamber and in the mandrel rotated to expose the sample to vacuum, then steam, and then vacuum again. The operation of the 2 platter valves was considered 2 axes of rotation and the operation of the mandrel a third axis of rotation. Full VSV treatment with the 2 platter valves and mandrel was called 3 axes treatment or 3 axes for short and VSV treatment with the mandrel only was called 1 axis treatment. With multiple cycles, the sequence of vacuum then steam was repeated multiple times. Process variables were mandrel design, carcass orientation within the chamber, source of chicken, full VSV treatment (called 3 axes of rotation), or mandrel VSV treatment only

(called 1 axis of rotation), steam temperature, steam time, initial, intermediate, and final vacuum times, and number of cycles. After treatment, the ball valve rotated 90 degrees to expose the sample to atmosphere. The sample was aseptically removed manually after treatment.

Sample Preparation

Chicken carcasses used for pilot plant experiments were purchased at local supermarkets. Chicken carcasses from the supermarkets had typical total aerobic plate counts $\leq$ 3 log cfu/mL. The carcasses were inoculated with *Listeria innocua* or *E. coli* K-12. *Listeria innocua* was chosen as an inoculum because it is nonpathogenic and has similar or higher thermal resistance compared to *Listeria monocytogenes* (Ryser and Marth 1999). *E. coli* K-12 was chosen as an inoculum because it is nonpathogenic and naturally occurring *E. coli* would be assayed in the field studies.

The carcasses were inoculated by immersion in a suspension of 10^6 cfu/mL *Listeria innocua* or *E. coli* K-12 for 10 min. The carcasses were allowed to drain in ambient conditions for 30 min before experimentation. Details for the preparation and inoculation of the chicken carcasses are given in Kozempel *et al.* (2001).

Carcasses used for the field studies were not inoculated. They were obtained after the final wash before chill at a local poultry processor and immediately brought to the experimental site in an insulated container. The carcasses were processed with no preparation or treatment. Forty-eight carcasses were sampled over a two day period. Three carcasses each for twelve treatments and 12 plant run controls were microbiologically analyzed for total aerobes, coliforms, generic *E. coli*, and *Campylobacter*. After treatment carcasses were placed into individual bags containing 100 mL of sterile water (pH 7.6) and subjected to the low volume whole carcass rinse (Cox *et al.* 1978) using an automated shaker sampler (Dickens and Cox 1985). The carcasses were removed from the shaker and allowed to drip for 30 s before the diluent was decanted into sterile specimen cups for transport to the micro lab for preparation of dilutions and plating procedures.

Microbiology Testing

Samples were generated using the whole bird rinse procedure. In pilot plant samples treated carcasses were placed in sterile plastic bags with 400 mL of Butterfield buffer solution (Difco Laboratories, Detroit, MI) and manually rinsed for 60 s (60 shakes). The rinses for *L. innocua* were appropriately diluted with sterile 1% peptone water and plated onto Tryptose Agar (Difco Laboratories) using a Spiral plater (Spiral Biotech, Bethesda, MD). The plates were incubated at 37C for one day. Colonies were counted and expressed as cfu/mL (colony

forming units/mL). Aliquots for *E. coli* K-12 were plated on tryptose agar plates using a spiral plater (Spiral Biotech, Cincinnati, OH) in duplicate and incubated overnight at 28C. Details of the microbiology assay procedures used for pilot plant samples are given in Kozempel *et al.* (2001).

In field trials, serial dilutions of the rinse diluent were prepared in peptone water. Total aerobic bacterial populations were enumerated on plate count agar (Difco, Detroit, MI). One tenth of a mL from a serial dilution of the rinse diluent was plated in duplicate on the surface of the agar, spread with a sterile bent glass rod, and incubated at 35C for 48 h prior to counting the colony forming units. *Campylobacter* were enumerated by plating in duplicate onto the surface of blood agar (Difco, Blaser). One tenth of a mL of a serial dilution of the rinse diluent was spread on the surface of each plate with a sterile bent glass rod, plates were then incubated at 42C for 36 to 48 h in a microaerophilic environment (5% O_2, 10% CO_2, and balance N_2). Following incubation colony forming units characteristic of *Campylobacter* were counted. All colonies counted as *Campylobacter* from each sample were confirmed as a member of the genus by examination of cellular morphology and motility on wet mount under phase contrast microscopy. Each colony type was further characterized as a member of the species *jejuni*, *coli*, or *lari* by a positive reaction on latex agglutination test kit (Integrated Diagnostics Inc., Baltimore, MD). Coliform and generic *E. coli* counts were made by plating 1 mL of the serial dilution from the rinse diluent onto duplicate *E. coli* petrifilm™ plates (3M Health Care, St. Paul, MN). Petrifilm™ plates were incubated at 35C for 48 h and colony types characteristic of coliform and *E. coli* were counted.

Experimental Designs

Experimental designs, 2^2, 2^3 and 2^4 factorial, (Davies *et al.* 1960) were used. Treatment samples generally consisted of three replicates. The data from the factorial designs were analyzed by analysis of variance using the replicate within treatment as error terms. When $P \leq 0.05$ the result was called statistically significant, at $P \leq 0.01$ the result was called highly statistically significant. Because of the potential for wide variation in results using a very inhomogeneous material like chicken carcasses, we included results at $P \leq 0.10$ and referred to them as possibly significant (Davies *et al.* 1961). Control samples of inoculated chicken carcasses were taken to provide an independent estimate of the extent of bacteria kill in the pilot plant research. Control samples of chicken carcasses using the native flora were taken to provide an independent estimate of the extent of bacteria kill in the field studies. A null hypothesis (Volk 1958) was made on the difference between means (H_o; $mean_1 = mean_2$) to compare the mean bacteria counts at various process conditions.

RESULTS AND DISCUSSION

Pilot Plant - I

Initial attempts using 1 cycle to process broilers resulted in extensive mechanical damage to the carcasses. The carcasses were ripped up the side and many actually fell off the mandrel for lack of sufficient integrity. These initial mandrels were nothing more than a perforated pipe of various lengths and diameters.

The side of the carcass is quite thin and easily ripped. The mandrels were changed to incorporate support for the cavity. Heavy gauge screening was added to support the carcass on the inside from the neck down. This eliminated the mechanical damage but resulted in less than 1 log kill of inoculated *L. innocua*. After trying 10 mandrel designs two were chosen for the field studies, mandrels designated IIIa and IVb. Mandrel IIIa was an open design with 3 curved rods approximating the shape of the cavity. The rods extended from the neck to a tray about half way down the carcass. Mandrel IVb was similar except there was no tray at the base. With these changes, there was still some unacceptable mechanical damage usually resulting in a small tear in the side. The mechanical damage was less frequent and less severe using mandrels IIIa and IVb than using the previous mandrels.

Table 1 summarizes the microbiology results for this basic design for the mandrel using full VSV treatment, 3 axes, (surface VSV treatment plus the mandrel VSV treatment). There was some tearing in 75% of the samples. The mechanical damage is unacceptable. There was no statistically significant difference in bacteria kill between 2 and 3 cycles or between any of the mandrels. Therefore, the data were pooled. The mean kill for inoculated *L. innocua* was 1.13 log cfu/mL (S.D. = 0.3488, n = 58).

The VSV process was run using only the mandrel, 1 axis, and not the two side platter valves. The mechanical damage was almost eliminated. Table 2 presents these results using only the mandrel, 1 axis, with no surface treatment from the other 2 axes. There was a statistically significant difference between the mandrels and between 2 and 3 cycles. The occurrence of mechanical damage was 12% and relatively mild. The mean bacteria kill for inoculated *L. innocua* using only the mandrel VSV treatment (1 axis, no surface treatment) was 0.87 log cfu/mL (S.D. = 0.2078, n = 17), Table 2. The mean kill for inoculated *L. innocua* using full VSV treatment, 3 axes, was 1.13 log cfu/mL (S.D. = 0.3488, n = 58), Table 1. Full VSV treatment (3 axes), 1.13 log kill, was highly statistically significantly better, $P = 0.01$, than mandrel VSV treatment alone (1 axis), 0.87 log kill.

TABLE 1.
PILOT PLANT-I MICROBIOLOGICAL RESULTS USING VARIOUS MANDRELS

		L innocua, log cfu/ml		
Mandrel	# Cycles	Control	Mean	Kill
Ib	2	6.07	5.06	1.01
IIa	2	6.07	5.19	0.88
IIa	2	5.93	4.87	1.06
III	2	5.92	4.61	1.31
IIIa	2	6.10	4.78	1.32
IIIa	3	6.10	4.94	1.16
IV	2	6.10	4.79	1.31
IV	3	6.10	5.11	0.99
IIIa	2	6.19	5.17	1.02
IIIa	2	6.11	4.90	1.21
IV	2	6.11	4.87	1.24
IVa	2	6.11	5.00	1.11
IVb	2	6.11	4.92	1.19

Mean kill = 1.13 log cfu/mL
S.D. = 0.3488
N = 58

TABLE 2.
PILOT PLANT-I MICROBIOLOGICAL RESULTS USING THE MANDREL ONLY

		L innocua, log cfu/ml		
Mandrel	# Cycles	Control	Mean	Kill
IIIa	2	6.19	5.34	0.85
IIIa	3	6.19	5.20	0.99
IV	2	6.19	5.21	0.98
IV	3	6.19	5.37	0.82
IIIa	2	6.11	5.33	0.78

Mean kill = 0.87 log cfu/mL
S.D. = 0.2078
N = 17

Two 2^3 factorial experimental designs were made using these 2 mandrels. These were the last 2 experiments before a scheduled field test using freshly killed chickens. *L. innocua* would not be determined in field tests. Field tests would determine *E. coli*, *Campylobacter*, coliforms, and APC. Therefore, for these 2 Pilot Plant experiments, the chicken carcasses were inoculated with nonpathogenic *E. coli* K-12. A total of four variables were studied, full VSV treatment (3 axes) or partial VSV treatment (1 axis), cycles, steam time, and the 2 different mandrels. The two 2^3 factorial experimental designs were combined into one 2^4 design shown in Table 3. The initial bacteria counts for the two experiments before treatment, based on the control samples, were virtually

TABLE 3.
2^4 EXPERIMENTAL DESIGN AND ANALYSIS OF VARIANCE FOR BACTERIAL KILL OF *E. COLI*-PILOT PLANT - I

Experimental Factors	Factor Levels	
	—	+
A	F	M
B	2	3
C	0.05	0.10
D	Ivb	IIIa

A = Treatment, full (F) or mandrel only (M)
B = # of cycles
C = steam time, s
D = Mandrel, IVb, IIIa
Initial and intermediate vacuum times = 0.1 s
Final vacuum time = 0.5 s

Experimental factors and interactions	*E. coli*			
	Mean, Log cfu/ml	Mean kill, Log cfu/ml	Mean Square	F value
A	4.31	1.25	0.1610	4.25*
B	4.15	1.41	0.1496	3.95$^+$
AB	4.26	1.30	0.0056	0.15
C	4.19	1.37	0.0310	0.82
AC	4.33	1.23	0.0061	0.16
BC	4.12	1.44	0.1452	3.83$^+$
ABC	4.30	1.26	0.0001	0.004
D	4.22	1.44	0.0280	0.74
AD	4.37	1.29	0.0016	0.04
BD	4.26	1.40	0.0331	0.87
ABD	4.47	1.19	0.0001	0.0002
CD	4.63	1.03	0.0408	1.08
ACD	4.64	1.02	0.0363	0.96
BCD	4.22	1.44	0.1704	4.50*
ABCD	4.26	1.40	0.0004	0.01
Error			0.0379	

Treatment samples consisted of three replicates.
Significant differences represented by; * $P \leq 0.05$, $^+P \leq 0.10$
Control (11 replicates) = 5.61 Log cfu/mL (S.D. = 0.1152)
Mean Square is the measure of variability attributed to the experimental factor or interaction.

identical (5.56 and 5.66 log cfu/mL). The pooled mean for the controls was 5.61 log cfu/mL (S.D. = 0.1152, n = 11). The bacteria count data could probably be used for analysis of variance but we chose to use kill data to remove any bias due to any difference in the initial bacteria counts. There was no statistically significant difference between steam times or between mandrels. There was a statistically significant difference ($P \leq 0.05$) between full VSV

treatment (3 axes) and partial VSV treatment (1 axis). There was a possible statistically significant difference ($P \leq 0.10$) between 2 of 3 cycles. The mean kill at full treatment and 2 cycles was 1.36 log cfu/mL. The mean kill using 3 cycles was also 1.36 log cfu/mL. Incidence of tearing was 10%, still unacceptable but much improved.

A Null Hypothesis was made to compare the kill using the VSV process for *E. coli* versus *L. innocua*. Using the full VSV treatment, 3 axes, the mean kill was 1.36 log cfu/mL for *E. coli* and 1.13 log cfu/mL for *L. innocua*. The kill for *E. coli* was statistically significantly greater ($P \leq 0.0002$). Using just the mandrel, 1 axis, the kill for *E. coli* (1.24 log cfu/mL) was statistically significantly ($P \leq 0.0001$) greater than for *L. innocua* (0.87 log cfu/mL). In both cases the kill was statistically significantly better for *E. coli*.

Field Tests

Although the mechanical damage problem was not solved at that time, the field tests were conducted as scheduled to determine microbiological kill. The VSV unit was installed on a flat bed truck and driven to the Richard Russell Research Center (RRRC) in Athens, GA. Freshly slaughtered and processed chickens were pulled before the chill tank and immediately transported unchilled to the RRRC.

A 2^3 factorial experimental design was made using steam time, cycles and full VSV treatment (3 axes) or partial VSV treatment (1 axis) as the independent variables. The responses were bacteria counts of *E. coli*, *Campylobacter*, coliforms, and total aerobic plate count (APC). The results are shown in Tables 4, 5, 6, and 7.

Table 4 presents the Analysis of Variance using *E. coli* count data. The only statistically significant independent variable ($P \leq 0.05$) was C, full VSV treatment (3 axes) or partial VSV treatment (1 axis). The mean kill was used to compare the results to the pilot plant results. The mean kill at the high level of C, full VSV treatment (3 axes), was 1.30 log cfu/mL (S.D. = 0.7251, n = 12). A Null Hypothesis revealed no statistically significant difference from the pilot plant results, 1.36 log cfu/mL.

The results using the mandrel VSV treatment only, 1 axis, were not as good as the pilot plant results. The kill was 0.84 log cfu/mL (S.D. = 0.6635, n = 12). A Null Hypothesis confirmed this was statistically significantly ($P \leq 0.001$) different from the pilot plant kill of 1.24 log cfu/mL.

Table 5 presents the Analysis of Variance for the *Campylobacter* count data. The only statistically significant independent variable ($P \leq 0.01$) was C, full VSV treatment (3 axes) or partial VSV treatment (1 axis). The mean kill for the high level of C, full VSV treatment (3 axes), was 1.23 log cfu/mL (S.D. = 0.3312, n = 12) which corresponds to a mean count of 1.43 log cfu/mL. There

was no statistically significant difference between the *E. coli* kill and *Campylobacter* kill (Null Hypothesis, $P = 0.2463$). These microbiology results confirmed our pilot plant research using nonpathogenic inoculated *Listeria innocua* and *E. coli* K-12.

TABLE 4.
2^3 EXPERIMENTAL DESIGN AND ANALYSIS OF VARIANCE FOR NATURALLY OCCURRING *E. COLI* - FIELD TEST # 1

Experimental Factors	Factor Levels	
	—	+
A	0.05	0.10
B	2	3
C	M	F

A = steam time, s
B = # of cycles
C = Treatment, full (F) or mandrel only (M)
Initial and intermediate vacuum times = 0.1 s
Final vacuum time = 0.5 s

Experimental Factors and Interactions	*E. coli*			
	Mean, Log cfu/ml	Mean Kill, Log cfu/ml	Mean square	F value
A	2.74	0.12	0.1247	0.44
B	2.42	0.44	0.3528	1.25
AB	1.35	1.51	1.1050	3.92
C	1.20	1.66	1.2560	4.45*
AC	1.17	1.69	0.0459	0.16
BC	1.69	1.17	1.5560	5.51*
ABC	2.18	0.68	2.8910	10.25**
Error			0.2822	

Treatment samples consisted of six replicates
Significant differences represented by; * $P \leq 0.05$, ** $P \leq 0.01$
Control (6 replicates) = 2.86 Log cfu/mL (S.D. = 0.8065)
Mean Square is the measure of variability attributed to the experimental factor or interaction

Table 6 gives the results for coliforms. The mean of the control samples was 2.99 log. The mean of the treated samples was 2.10 log and the kill was 0.9 log. This was a statistically significant ($P \leq 0.05$) kill. No single independent variable was statistically significant.

Table 7 gives the results for APC. No variable was statistically significant. The mean of the control samples was 3.36 log. The mean of the treated samples was 2.69 log. The kill was 0.7 log. This was a statistically significant ($P \leq 0.05$) kill. The aerobes were unidentified. They could be thermally sensitive or resistant.

TABLE 5.
2^3 EXPERIMENTAL DESIGN AND ANALYSIS OF VARIANCE FOR NATURALLY OCCURRING *CAMPYLOBACTER* – FIELD TEST # 1

Experimental Factors	Factor Levels	
	–	+
A	0.05	0.10
B	2	3
C	M	F

A = steam time, s
B = # of cycles
C = Treatment, full (F) or mandrel only (M)
Initial and intermediate vacuum times = 0.1 s
Final vacuum time = 0.5 s

Experimental Factors and Interactions	*Campylobacter*			
	Mean, Log cfu/ml	Mean Kill, Log cfu/ml	Mean square	F value
A	2.01	0.65	0.0088	0.06
B	1.69	0.97	0.0400	0.26
AB	2.17	0.49	0.0748	4.89*
C	1.70	0.96	1.5500	10.13**
AC	1.30	1.36	0.6468	4.22
BC	1.53	1.13	0.0160	0.10
ABC	1.20	1.46	0.0368	0.24
Error			0.1531	

Treatment samples consisted of six replicates
Significant differences represented by; * $P \leq 0.05$, ** $P \leq 0.01$
Control (6 replicates) = 2.66 Log cfu/mL (S.D. = 0.5001)
Mean Square is the measure of variability attributed to the experimental factor or interaction

As expected from the pilot plant research, there was mechanical damage. The carcasses used in this field test were much smaller breasted than the ones used in the pilot plant studies. The mechanical damage was greater than expected because of the difference in the carcass dimensions. Mandrel IVb is very open and permits the carcass to hang low in the chamber. Large breasted carcasses, used in the pilot plant study had much less damage than the small breasted chickens that sat lower in the chamber. When the main valve closed, one of the legs invariably got stuck in the valve, breaking the end of the leg at the hock. This caused two problems, a vacuum and steam leak and mechanical damage. When the leg got caught, it pulled away from the carcass side and ripped the side open. The same thing happened in the pilot plant with earlier mandrels.

The exposed meat surface exhibited some minor thermal damage. A thin white film formed on the exposed surface. Some of the film can be wiped off. Much of the thermal damage dissipates over time and during cooling. The unexposed meat on the same sample was virtually unaffected.

TABLE 6.
2^3 EXPERIMENTAL DESIGN AND ANALYSIS OF VARIANCE FOR NATURALLY OCCURRING COLIFORM – FIELD TEST # 1

Experimental Factors	Factor Levels	
	–	+
A	0.05	0.10
B	2	3
C	M	F

A = steam time, s
B = # of cycles
C = Treatment, full (F) or mandrel only (M)
Initial and intermediate vacuum times = 0.1 s
Final vacuum time = 0.5 s

Experimental Factors and Interactions	Coliform			
	Mean, Log cfu/ml	Mean Kill, Log cfu/ml	Mean square	F value
A	2.55	0.44	0.0704	0.15
B	2.52	0.47	0.3601	0.78
AB	1.61	1.38	0.2817	0.61
C	1.95	1.04	0.0020	0.004
AC	1.72	1.27	0.0913	0.20
BC	2.03	0.96	0.5163	1.11
ABC	2.72	0.27	2.7340	5.89*
Error			0.4641	

Treatment samples consisted of six replicates
Significant differences represented by; * $P \leq 0.05$, ** $P \leq 0.01$
Control (6 replicates) = 2.99 Log cfu/mL (S.D. = 0.7077)
Mean Square is the measure of variability attributed to the experimental factor or interaction

There was one unexpected result. Hard scald chickens were used during the field test in Athens. Hard scald is done at 58C and removes the epidermal layer. During pilot plant studies only soft scald chickens (52C) were used. When processed, the skin on the soft scald carcasses retained or increased the yellow color. When the hard scald carcasses were processed, the skin suffered some thermal damage, exhibiting a pale yellow color. This could be a problem in a consumer market that uses the hard scald chicken.

There is a difference of opinion among the authors on the effect of the VSV process on hard scald carcasses. One author is of the opinion that the yellowing of the skin would preclude sale in the fresh chicken market. Others knowledgeable in the poultry industry think the thermal damage is too slight to be important; that in fact, the slight change may be an asset indicating to the consumer that the chicken has been nonchemically treated for safety. The skin on soft scald carcasses appears to be unchanged except for a little shrinkage.

TABLE 7.
2^3 EXPERIMENTAL DESIGN AND ANALYSIS OF VARIANCE FOR NATURALLY OCCURRING TOTAL AEROBIC PLATE COUNT (APC) - FIELD TEST # 1

Experimental Factors	Factor Levels	
	—	+
A	0.05	0.10
B	2	3
C	M	F

A = steam time, s
B = # of cycles
C = Treatment, full (F) or mandrel only (M)
Initial and intermediate vacuum times = 0.1 s
Final vacuum time = 0.5 s

Experimental Factors and Interactions	APC			
	Mean, Log cfu/ml	Mean Kill, Log cfu/ml	Mean square	F value
A	2.71	0.65	0.0459	0.11
B	2.71	0.65	0.4510	1.09
AB	1.96	1.40	0.0260	0.06
C	3.03	0.33	0.8251	2.00
AC	2.93	0.43	0.3927	0.95
BC	2.56	0.80	0.0315	0.08
ABC	3.00	0.36	0.6970	1.69
Error			0.4136	

Treatment samples consisted of six replicates
Control (6 replicates) = 3.36 Log cfu/mL (S.D. = 0.6290)
Mean Square is the measure of variability attributed to the experimental factor or interaction

Much of the chicken sold is as breaded chicken parts, whether sold directly to the consumer or through restaurants or fast food establishments. Whether the VSV process will interfere with proper batter sticking is an unanswered question. We think the experts on poultry — the poultry industry itself; best address this and other organoleptic and quality issues.

A second field test was made using a 2^2 factorial experimental design with steam time and number of cycles as the independent variables. The second field test used full VSV treatment, 3 axes. These carcasses had much lower bacteria counts. *Campylobacter* was too low to do an Analysis of Variance. Four of 6 controls tested positive for *Campylobacter* (1.0, 1.3, 1.6, 1.9 log cfu/mL) but only 1 of 12 treated samples tested positive (1.0 log cfu/mL). Table 8 presents the Analysis of Variance for *E. coli*. No variable was statistically significant. The mean count of treated carcasses was less than 1 log cfu/mL (0.59 log, S.D. = 0.5125). The mean kill was 1.0 log cfu/mL.

TABLE 8.
2^3 EXPERIMENTAL DESIGN AND ANALYSIS OF VARIANCE FOR NATURALLY OCCURRING *E. COLI* - FIELD TEST # 2

Experimental Factors	Factor Levels	
	−	+
A	0.05	0.10
B	2	3

A = steam time, s
B = # of cycles
Treatment, full (F)
Initial and intermediate vacuum times = 0.1 s
Final vacuum time = 0.5 s

Experimental Factors and Interactions	*E. coli*			
	Mean, Log cfu/ml	Mean Kill, Log cfu/ml	Mean square	F value
A	0.99	0.56	0.0494	0.18
B	0.47	1.08	0.4602	1.72
AB	0.32	1.23	0.2380	0.89
Error			0.2678	

Treatment samples consisted of six replicates
Control (6 replicates) = 1.55 Log cfu/mL (S.D. = 0.1178)
Mean Square is the measure of variability attributed to the experimental factor or interact

Table 9 presents the Analysis of Variance for coliform. No variable was statistically significant. The mean count of treated carcasses was 0.62 log cfu/mL (S.D. = 0.5397). The mean kill was 1.1 log cfu/mL. Table 10 presents the Analysis of Variance for APC. Analysis of variance indicated number of cycles was possibly statistical significant ($P \leq 0.10$). The mean count of treated carcasses was 2.56 log cfu/mL (S.D. = 0.5920). The mean kill was 0.8 log cfu/mL.

Mandrel IIIa was used in these tests because it was designed with more support for the internal cavity of the carcass, but was too big for the carcasses used in this test. It was a modification of an earlier mandrel. It works fine with large breasted birds with large visceral cavities. But, it is very difficult to put the carcass on the mandrel if the cavity is small. For this mandrel the cavity must be larger than 7.5 cm. In the second field test the cavities were too small for the mandrel. The carcasses did not fit. They had to be forced on and many ripped.

TABLE 9.
2^3 EXPERIMENTAL DESIGN AND ANALYSIS OF VARIANCE FOR NATURALLY OCCURRING COLIFORM – FIELD TEST # 2

Experimental Factors	Factor Levels	
	—	+
A	0.05	0.10
B	2	3

A = steam time, s
B = # of cycles
Treatment, full (F)
Initial and intermediate vacuum times = 0.1 s
Final vacuum time = 0.5 s

Experimental Factors and Interactions	Coliform			
	Mean, Log cfu/ml	Mean Kill, Log cfu/ml	Mean square	F value
A	1.01	0.71	0.0494	0.16
B	0.48	1.24	0.4840	1.57
AB	0.35	1.37	0.2107	0.69
Error			0.3075	

Treatment samples consisted of six replicates
Significant differences represented by; $^{+}$ $P \leq 0.10$, ** $P \leq 0.01$
Control (6 replicates) = 1.72 Log cfu/mL (S.D. = 0.1624)
Mean Square is the measure of variability attributed to the experimental factor or interaction

TABLE 10.
2^3 EXPERIMENTAL DESIGN AND ANALYSIS OF VARIANCE FOR NATURALLY OCCURRING TOTAL AEROBIC PLATE COUNT (APC) – FIELD TEST # 2

Experimental Factors	Factor Levels	
	—	+
A	0.05	0.10
B	2	3

A = steam time, s
B = # of cycles
Treatment, full (F)
Initial and intermediate vacuum times = 0.1 s
Final vacuum time = 0.5 s

Experimental Factors and Interactions	APC			
	Mean, Log cfu/ml	Mean Kill, Log cfu/ml	Mean square	F value
A	3.15	0.20	0.0140	0.06
B	2.45	0.90	1.1970	4.94^{+}
AB	2.04	1.31	0.7057	2.91
Error				

Treatment samples consisted of six replicates
Control (6 replicates) = 3.35 Log cfu/mL (S.D. = 0.3933)
Mean Square is the measure of variability attributed to the experimental factor or interaction

Pilot Plant - II

Since the field studies duplicated or exceeded the microbiological kill found in the pilot plant, the objective of the follow-up pilot plant studies was to eliminate the mechanical damage experienced in the field while maintaining the microbiological kill. The leg breakage was simple to eliminate. The previous prototype processor had a screen in the bottom to hold the chicken. Once the mandrel was used to support the carcass, it was considered superfluous. However, in retrospect, it kept the legs from extending beyond the chamber, therefore a screen was added to the bottom of mandrel IVb to support the legs. Leg breakage was no longer a problem.

A consequence of adding the screen was that some carcasses, especially ones with long necks, sat too high in the chamber. When the valve closed the combination of rubbing of the inside of the valve and the steam pressure within the cavity caused some skin ripping and splitting of the neck area. Lowering the neck support position on the mandrel corrected the neck abrasion problem. Depending on the size of the cavity, the steam still caused some damage because there was too much steam applied to the cavity. The inside diameter of the mandrel pipe was 21 mm. A constrictor sleeve was added to the mandrel pipe, where the steam exited into the carcass cavity, that reduced the inside diameter to 12 mm. This solved the damage problem due to excess steam in the neck area.

Ten 2^3 factorial experimental designs were made during the mechanical damage study evaluating the effect of such factors as carcass orientation, carcass height, and mandrel pipe diameter on the mechanical damage. These experiments used full VSV treatment (3 axes). The effect of these 3 factors plus independent variables cycles (2 or 3), steam temperature (127 or 138C), and source or brand of chicken on microbiological kill of inoculated *E. coli* was evaluated. The mechanical modifications had no statistically significant effect on the microbiological kill. There was no statistically significant difference between steam temperatures.

Table 11 summarizes the results of these experiments categorized by source and number of cycles. The overall mean kill was 1.2 log cfu/mL, SD = 0.3112, n = 216). Statistically three cycles were significantly better than 2 cycles. There were statistically significant differences associated with chicken source. Source A at 3 cycles was statistically significantly different ($P \leq 0.001$) than the other combinations of source of chicken and number of cycles and significantly better ($P \leq 0.001$) than the same source at 2 cycles. The mean kill for source A at 3 cycles was 1.49 log cfu/mL, SD = 0.2137, n = 36 and at 2 cycles the log kill was 1.24, SD = 0.2664, n = 36. There was a statistically significant difference between sources B and C at 3 cycles and between B and C at 2 cycles. The kill for source B at 3 cycles was 1.24 (SD = 0.2595, n =

36) and for source C was 1.08 (SD = 0.2912, n = 36). The kill for source B at 2 cycles was 1.14 (SD = 0.2289, n = 36) and for source C was 0.97 (SD = 0.3346, n = 36).

TABLE 11.
BACTERIA KILL OF INOCULATED *E. COLI* K-12 - PILOT PLANT-II

	E. coli K-12, log cfu/ml (SD)	
	2 cycles	3 cycles
Source A	1.24 (0.2664)	1.49 (0.2137)
Source B	1.14 (0.2289)	1.24 (0.2595)
Source C	0.97 (0.3347)	1.08 (0.2912)

We have no definitive explanation for the difference in effectiveness of treatment of carcasses from different sources. Source A was hard scald, organically raised chickens. They were somewhat smaller carcasses than the other two sources, averaging 1400 - 1800 g. The giblets were packed in paper in the cavity. These carcasses were bulk shipped packed in ice.

Source B and C were soft scald, conventionally raised carcasses that averaged 1600 – 2000 g. The carcasses were individually packed in plastic film. The giblets were packed loose inside the cavity of source C and packed in plastic in the cavity in source B carcasses. The giblets were removed from all carcasses before processing. Commercially, the VSV process would be used before the giblets would be inserted.

There are certainly numerous other differences among the sources that are unknown to the authors and which may be the causative factor or factors for the difference in effectiveness of treatment. It could be the genetics of the flocks used or as simple as feed ingredients. It is probably a combination of factors.

The chicken cavity is an impediment to treatment. Specific mandrels can be designed to treat each size and type of carcass or a malleable mandrel could be developed that would fit all carcasses within reasonable tolerances. Although we have been successful in treating whole chicken carcasses, chicken parts are much simpler to process and usually give somewhat better bacteria kill.

Chicken quarters (leg plus thigh) were processed to determine the kill possible on chicken parts that include no cavity. The quarters had no cavity and therefore required no mandrel to process. Sources B and C were used. The kill of inoculated *E. coli* K-12 on the quarters was 1.73 log cfu/mL, SD = 0.1121, n = 5 which was statistically significantly better than for the sources B and C whole carcasses using 3 cycles (1.24 and 1.08 log kill, respectively).

The essential elements of the process are rapid application of vacuum and steam. The VSV processor used in these studies is a prototype. Although the

carcasses were treated in about 1 s depending on the number of cycles, this does not include the time to feed and withdraw the product. The exact design and fabrication of a commercial unit will be the province of industry, although we anticipate cooperating in the effort.

The design of the unit might depend on the location in the process. Initially, we thought the VSV unit would be installed after the chill tank. In this location, the process could be integrated into the packaging step. There is sufficient time on a packaging conveyor to treat the carcass before sealing.

Industry has indicated that it might be preferable to install the VSV unit before the chill tank. In this case, a commercial unit would probably consist of multiple chambers or multiple units instead of a single chamber to eliminate the time required for feeding and ejecting a carcass. Feed would be automatic, not manual as in these studies. A conveyor would be integrated into the unit to synchronize feeding the carcass with the opening of the main chamber valve. The exiting carcass would simply drop into the chill tank.

Of course, these processing steps could add some time to the overall process; but, as long as the time to treat is at the same or faster rate as the rest of the process, the VSV process would not affect line speed.

CONCLUSIONS

The field VSV surface intervention process killed 1.4 log of *E. coli* and 1.2 log of *Campylobacter* on freshly processed chicken using 3 cycles and 138C saturated steam. The same process in the pilot plant killed 1.1-1.5 log of inoculated *E. coli* K-12 depending on the source of chicken. The total process time was 1.1 s.

Originally, it was hoped to achieve a bacteria kill of 5 logs on chicken carcasses to achieve pasteurization. It is apparent that there are inherent difficulties associated with poultry that limit the bacteria kill in this prototype to less than 2 logs even though much higher kills have been realized with the VSV process on other products, for example 5 log kill on hot dogs (Kozempel *et al.* 2000a). As the only hurdle in a poultry plant, it would not be sufficient. However, in chicken processing plants, the bacteria are killed in a series of 0.5 - 1 log kill steps (hurdles) such as in the scald tanks, vent operations, inside out washers, chlorine spray cabinets, and the chlorinated chill tanks (private communications). No single step or hurdle does it all. Therefore, the VSV process for chicken should be considered one component of a hurdle technology and not as a stand-alone pasteurizer. As part of a hurdle technology, processes that achieve a 1 – 1.5 log kill should be of interest.

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